

**M<sub>3</sub> MUSCARINIC RECEPTOR, HUMAN RECOMBINANT**

Expressed in Sf9 Cells

Product Number **M-176****Product Information****Product Description**

M<sub>3</sub> Muscarinic Receptor, human recombinant (Sf9) is a frozen aliquot of membranes from Sf9 cells transfected with the human recombinant M<sub>3</sub> muscarinic receptor.

**Reagent**

M<sub>3</sub> Muscarinic Receptor, human recombinant (Sf9) is suspended in 50 mM Tris-HCl, pH 7.4, 10 % glycerol, and 1 % bovine serum albumin (BSA).

**Procedure***Incubation buffer:*

50 mM Tris-HCl, pH 7.4  
10 mM MgCl<sub>2</sub>  
1 mM EDTA

*Binding Protocol*Membranes:

Dilute in incubation buffer (0.5 mL of membrane + 24.5 mL incubation buffer).

Assay mixture:

500  $\mu$ L diluted membranes  
20  $\mu$ L radioligand  
20  $\mu$ L buffer or unlabeled ligand

Radioligand:

[<sup>3</sup>H]-N-Methyl scopolamine at a final concentration of 0.1 nM for competition studies.

Unlabeled ligand:

Atropine at a concentration of 1.0  $\mu$ M.

Incubation time:

60 minutes at 27 °C

Separation:

Over GF/C filter (5 mm diam., presoaked in 0.3 % polyethylamine prepared in incubation buffer) then washed 4x with 500  $\mu$ L of ice cold 50 mM Tris-HCl, pH 7.4.

**Results**

Typical affinities using standard binding assay above. Results may vary from lot to lot.

Ligand	K <sub>i</sub> (nM)
[ <sup>3</sup> H]-N-Methyl scopolamine	0.19 (Kd)
4-DAMP methiodide	0.87
Pirenzepine	28

**Storage/Stability**

Store tightly sealed at -80 °C. The membranes will retain their original specific activity for several months when stored at -80 °C in the original packing solution. Repeated freeze-thaw of this product is not recommended.

**Precautions**

While no human toxicity data is available for this substance, it should be handled with care. Precautions should be taken to avoid contact by all routes of exposure.

**References**

1. Tobin, A.B. and Nahorski, S.R., *J. Biol. Chem.*, **268**, 9817 (1993).
2. Wall, S.J. et al., *Mol. Pharmacol.*, **40**, 783 (1991).

mje 04/01

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